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I, LEANNE MYNOTT, TEAM LEADER EXAMINATION SUPPORT AND SALES hereby certify that annexed is a true copy of the Provisional specification in connection with Application No. PP 8685 for a patent by NOVOGEN RESEARCH PTY LTD filed on 15 February 1999.

WITNESS my hand this
Ninth day of March 2000

LEANNE MYNOTT
TEAM LEADER EXAMINATION
SUPPORT AND SALES

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PROVISIONAL SPECIFICATION

Applicant(s): Novogen Research Pty Ltd
140 Wicks Road
North Ryde New South Wales 2113
AUSTRALIA

Address for Service: DAVIES COLLISON CAVE
Patent & Trade Mark Attorneys
Level 10, 10 Barrack Street
SYDNEY NSW 2000

Invention Title: **Production of isoflavone derivatives.**

The invention is described in the following statement:

PRODUCTION OF ISOFLAVONE DERIVATIVES

Introduction

The present invention relates to the hydrogenation of isoflavones and products thereof. The invention also relates to the synthesis of phytoestrogenic isoflavone metabolites from the hydrogenation products of isoflavones.

Background of the Invention

Isoflavone metabolites possess a very wide range of important biological properties including oestrogenic effects (WO 98/08503). Isoflavone metabolites can be isolated from the urine of human volunteers subjected to diets rich in plant isoflavanoids such as soya, lentils, peas and beans.

In spite of the recently discovered biological significance of isoflavone metabolites there is not at present a general method suitable for the large scale synthesis of these metabolites. The few reported syntheses of these metabolites utilise either catalytic hydrogenation or hydrogen transfer reduction of the corresponding isoflavones.

These reduction reactions are non-selective, extremely difficult to control and lead to mixtures of different products. Chromatography is required to separate the reaction products and gives low yields of isoflavanones, isoflavan-4-ols, isoflavenes and isoflavans. The chromatography required is tedious and often impractable for large scale reactions. Furthermore, attempts to improve the yield and purity of products obtained from hydrogenation reactions has been met with limited success as evidenced by published results which are largely contradictory.

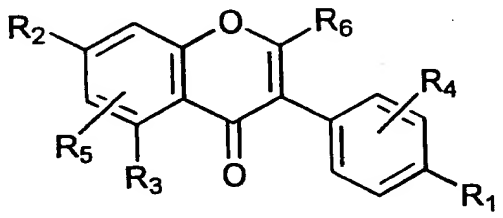
Known solvents used in dehydrogenation reactions include N-methylpyrrolidinone which is a severe eye irritant and a possible carcinogen. The high boiling point of N-methylpyrrolidinone makes it extremely difficult to remove after the reduction.

Isoflavan-4-ols are key intermediates in the synthesis of isoflavenes and accordingly there is a need for a more efficient and reliable synthesis of isoflavan-4-ols. Therefore it is an object of the present invention to overcome or at least alleviate one or more of the above-mentioned disadvantages of the prior art.

Surprisingly conditions have been found by the present inventors which enable the hydrogenation of isoflavones to afford excellent yields of relatively pure tetrahydroisoflavanol products.

Summary of the Invention

Thus the present invention provides a method for the hydrogenation of a compound of formula I



(I)

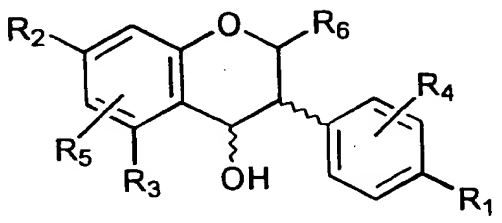
wherein

R_1 and R_2 are independently OR_7, $OC(O)R_7$, $OS(O)R_7$, alkyl, aryl, arylalkyl, thio, alkylthio, amino, alkylamino, dialkylamino, nitro, or halo,

R_3, R_4, R_5 and R_6 are independently hydrogen, hydroxy or R_1, and

R_7 is alkyl, fluoroalkyl, aryl or arylalkyl,

to prepare a compound of formula II

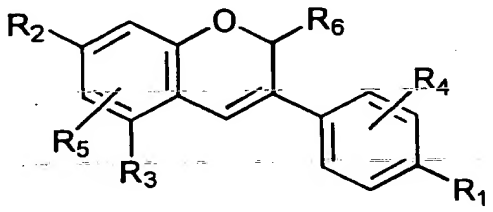


(II)

wherein

R_1, R_2, R_3, R_4, R_5, R_6 and R_7 are as defined above.

The present invention also provides a method for the dehydration of a compound of formula II, which method may optionally include deprotection or transformation steps, to prepare a compound of the formula III



(iii)

wherein

R_1 and R_2 are independently hydroxy, OR_7, $OC(O)R_7$, $OS(O)R_7$, alkyl, aryl, arylalkyl, thio, alkylthio, amino, alkylamino, dialkylamino, nitro, or halo,

R_3, R_4, R_5 and R_6 are independently hydrogen, hydroxy or R_1, and

R_7 is alkyl, fluoroalkyl, aryl or arylalkyl.

The present invention also provides a compound of formula II or III when prepared by a method described above and pharmaceutical compositions comprising same.

Detailed Description of the Invention

The starting isoflavone of formula I, the isoflavan-4-ol of formula II and the isoflavene of formula III preferably have the following substituents wherein

R_1 and R_2 are independently OR_7, $OC(O)R_7$, $OS(O)R_7$, thio, alkylthio, bromo, chloro or fluoro,

R_3, R_4, R_5 and R_6 are independently hydrogen or R_1, and

R_7 is alkyl or fluoroalkyl;

more preferably they have the following substituents wherein

R_1 and R_2 are independently OR_7 or $OC(O)R_7$,

R_3, R_4, R_5 and R_6 are hydrogen, and

R_7 is methyl, ethyl, propyl, isopropyl or trifluoromethyl;

and most preferably they have the following substituents wherein

R_1 and R_2 are independently OR_7 or $OC(O)R_7$,

R_3, R_4, R_5 and R_6 are hydrogen, and

R_7 is methyl or trifluoromethyl.

The particularly preferred compounds of formula I are 4',7-diacetoxyisoflavanone (daidzein diacetate) and 7-acetoxy-4'-methoxyisoflavone; the particularly preferred compounds of formula II are 4',7-diacetoxyisoflavan-4-ol (tetrahydrodaidzein diacetate) and 7-acetoxy-4'-methoxyisoflavan-4-ol; and the particularly preferred compounds of formula III are 4',7-diacetoxyisoflav-3-ene (dehydroequol diacetate), 4',7-dihydroxyisoflav-3-ene (dehydroequol), 7-acetoxy-4'-methoxyisoflav-3-ene and 7-hydroxy-4'-methoxyisoflav-3-ene.

The term "alkyl" is taken to mean both straight chain and branched chain alkyl groups such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, secbutyl, tertiary butyl, and the like. Preferably the alkyl group is a lower alkyl of 1 to 6 carbon atoms. The alkyl group may optionally be substituted by one or more of fluorine, chlorine, bromine, iodine, carboxyl, C_1-C_4-alkoxycarbonyl, C_1-C_4-alkylamino-carbonyl, di-(C_1-C_4-alkyl)-amino-carbonyl, hydroxyl, C_1-C_4-alkoxy, formyloxy, C_1-C_4-alkyl-carbonyloxy, C_1-C_4-alkylthio, C_3-C_6-cylcoalkyl or phenyl.

The term "aryl" is taken to include phenyl and naphthyl and may be optionally substituted by one or more C_1-C_4-alkyl, hydroxy, C_1-C_4-alkoxy, carbonyl, C_1-C_4-alkoxycarbonyl , C_1-C_4-alkylcarbonyloxy or halo.

The hydrogenation is ideally preformed with hydrogen in the presence of a reduction catalyst and a solvent. The reaction is preferably conducted under hydrogen at a pressure of 1-20 atmosphere, more preferably 1-5 atmospheres. The reaction may be performed from 15 to 60°C and is typically carried out at room temperature.

The reaction time may range from 12 hours to 72 hours or more and is typically about 55 hours or more. Generally better yields and cleaner reactions are achieved with longer reaction times. It will be appreciated that reaction conditions may be varied depending on the individual nature of the compounds and the progress of the hydrogenation reaction.

The reduction catalysts may be selected from heterogeneous catalysts (whereby the catalyst is insoluble in the reaction medium) or homogenous catalysts (whereby the catalyst is soluble in the reaction medium). Examples of heterogeneous reduction catalysts include Raney nickel, palladium black, palladium hydroxide on carbon, palladium on activated carbon (1% Pd to 30% Pd), palladium on alumina powder, palladium on various barium salts, sodium borohydride reduced nickel, platinum metal, platinum black, platinum on activated carbon (1% Pt to 10% Pt), platinum oxide, rhodium salts, ruthenium salts and their chiral salts and zinc oxide. Preferably the catalyst is palladium on activated carbon (1% Pd to 10% Pd), more preferably about 5% palladium on carbon.

Examples of homogeneous reduction catalysts include chlorotris(triphenylphosphine)rhodium, chloro(trisphenylphosphine)hydridoruthenium (II) and pentacyanocobaltate (II).

The solvents suitable for use in the present invention include but are not limited to C_1-C_8 alcohols and polyols, alkyl acetates, tetrahydrofuran, ethers, dioxane and C_1-C_3 acids. Preferably the solvent is a C_1-C_6 alcohol or C_1-C_6 alkyl acetate, more preferably ethanol, as well as methanol, propanol, isopropanol, butanol, isobutanol, secbutanol or tertiary butanol, and most preferably absolute ethanol.

diacetoxytetrahydrodaidzein (50% yield: 73% purity) (*cis* isomer 27%). Subsequent recrystallisations from ethanol afforded the pure *trans* isomer in 25% overall yield.

Likewise the 7-acetoxy-4'-methoxyisoflavan-4-ol was able to be fractionally recrystallised to give the pure *trans*-isomer.

Synthesis of tetrahydrodaidzein was achieved by removal of the protecting acetoxy groups under mild conditions using imidazole in ethanol at reflux. The tetrahydrodaidzein was isolated in 80% yield after crystallisation from aqueous ethanol.

Dehydration of the mixture of alcohols using benzoyl chloride/dimethylformamide at 100°C has been reported in the literature by Liepa to give the desired isoflavene. However this reaction could only be repeated in low yield. Dehydration may also be effected by treatment with acids such as sulfuric acid, hydrochloric acid, polyphosphoric acid, thionyl chloride and the like. Alternative methods of dehydration using *p*-toluenesulfonic acid and trifluoroacetic acid in refluxing dichloromethane were also investigated.

The dehydration reaction using *p*-toluenesulfonic acid gave low yields of product. Use of trifluoroacetic acid led initially to the formation of trifluoroacetate esters which upon prolonged heating gave the isoflavene but also in low yield.

Synthesis of dehydroequol was achieved by removal of the protecting acetoxy groups under mild conditions as described for the synthesis of tetrahydrodaidzein, and dehydroequol was purified by crystallisation from methanol/benzene.

The surprising results obtained by the present inventors are in sharp contrast to those reported in the literature for other attempted hydrogenations. One such marked advantage is the use of alcohol solvents such as absolute ethanol in the hydrogenation of isoflavanones. The isoflavanols prepared by the methods of the present invention are very crystalline and can be isolated in good purity without the need for chromatography. The

isoflavanols protected as esters can be readily deprotected under mild conditions. The isoflavanols can also be converted to isoflavenes by dehydration.

Throughout this specification and the claims which follow, unless the context requires otherwise, the word "comprise", and variations such as "comprises" or "comprising", will be understood to imply the inclusion of a stated integer or step or group of integers or steps but not the exclusion of any other integer or step or group of integers or steps.

The invention is further described in and illustrated by the following Examples. The Examples are not to be construed as limiting the invention in any way.

EXAMPLE 1

Synthesis of 4',7 Diacetoxydaidzein

Method A

A mixture of daidzein (1.0g, 3.9 mmol), acetic anhydride (5 ml) and pyridine (5 ml) was left in the dark at room temperature for 24 h. The reaction mixture was poured into water (100 ml), stirred for 2h and then extracted with dichloromethane (3 x 50 ml). The dichloromethane layer was washed with water, dried over anhydrous sodium sulfate and evaporated. The white residue was crystallised from methanol to yield daidzein diacetate as white prisms (1.1 g, 83%). ^{1}H NMR ($CDCl_3$): δ 2.32 (s, 3H, $OCOCH_3$), 2.36 (s, 3H, $OCOCH_3$), 7.18 (d, 2H, J 9.2 Hz, ArH), 7.19 (d, 1H, J 9.0 Hz, H6), 7.31 (d, 1H, J 2.0 Hz H8), 7.59 (d, 2H, J 9.2 Hz, ArH), 8.00 (s, 1H, H2), 8.33 (d, 2H, J 8.2 Hz, ArH).

Method B

A mixture of daidzein (2.0 g, 7.9 mmol), acetic anhydride (10 ml) and pyridine (2 ml) was heated on an oil bath at 105-110°C for 1h. After cooling the mixture to room temperature, it was stirred for a further 30 min during which time the diacetate crystallised from the solution. The product was filtered, washed thoroughly with water and recrystallised from methanol to yield daidzein diacetate as colourless prisms (2.4 g, 90%).

EXAMPLE 2

Synthesis of 7-acetoxy-4'-methoxyisoflavanone

A mixture of 7-acetoxy-4'-methoxyisoflavanone (2.0g, 7.5 mmol), acetic anhydride (10 ml) and pyridine (2 ml) was heated on an oil bath at 105-110°C for 1 hour. After cooling the mixture to room temperature, it was poured into water (100 ml), stirred for 2 hours and then extracted with dichloromethane (3 x 50 ml). The dichloromethane layer was washed with water, dried over anhydrous sodium sulfate and evaporated. The white residue was crystallised from methanol to yield 7-acetoxy-4'-methoxyisoflavanone as colourless prisms (2.1g, 91%). 1H NMR ($CDCl_3$): d 2.36 (s, 3H, $OCOCH_3$), 3.84 (s, 3H, OCH_3), 6.98 (d, 2H, J 8.7 Hz, ArH), 7.16 (dd, 1H, J 1.9 Hz 8.6 Hz, H6), 7.30 (d, 1H, J 1.9 Hz H8), 7.50 (d, 2H, J 8.7 Hz, ArH), 8.00 (s, 1H, H2), 8.32 (d, 1H, J 8.6 Hz, H5).

EXAMPLE 3

Synthesis of 4',7-diacetoxytetrahydrodaidzein (4'7-Diacetoxyisoflavan-4-ol)

Palladium-on-charcoal (5%, 0.08 g) was added to a suspension of 4',7-diacetoxydaidzein (0.5 g, 1.5 mmol) in absolute ethanol (400 ml) and the mixture was stirred at room temperature under a hydrogen atmosphere for 55 hours. The catalyst was removed by filtration through Celite and the filtrate was evaporated *in vacuo* to yield 4',7-diacetoxytetrahydrodaidzein (0.51 g, 100%) in quantitative yield. A nuclear magnetic resonance spectrum revealed the product to be a clean 1 : 1 mixture of cis and trans 4',7-diacetoxytetrahydrodaidzein.

trans-4',7-Diacetoxyisoflavan-4-ol; ^1H NMR ($CDCl_3$): δ 2.28 (s, 3H, $OCOCH_3$), 2.29 (s, 3H $OCOCH_3$), 3.14 (ddd, 1H, J 3.7 Hz, 7.9 Hz, 9.1 Hz, H3), 4.24 (dd, 1H, J 9.1 Hz, 11.3 Hz, H2); 4.35 (dd, 1H, J 3.7 Hz, 11.3 Hz, H2), 4.87 (d, 1H, J 7.9 Hz, H4), 6.61 (d, 1H, J 2.3 Hz, H8), 6.70 (dd, 1H, J 2.3 Hz, 8.4 Hz, H6), 7.06 (d, 2H, J 8.6 Hz, ArH), 7.23 (d, 2H, J 8.4 Hz, ArH), 7.44 (dd, 1H, J 0.8 Hz, 8.4 Hz, H5). ^{13}C NMR ($CDCl_3$): δ 20.98 ($OCOCH_3$), 46.18 (C3), 68.04 (C2), 69.01 (C4), 109.67 (C8), 114.26 (C6), 121.96, 128.96 (ArCH), 129.40 (C5).

cis-4',7-Diacetoxyisoflavan-4-ol: ^{1}H NMR ($CDCl_3$): δ 2.28 (s, 3H, $OCOCH_3$), 2.29 (s, 3H, $OCOCH_3$), 3.30 (dt, 1H, J 3.4 Hz, J 11.8 Hz, H3), 4.31 (ddd, 1H, J 1.4 Hz, 3.6 Hz, 10.5 Hz, H2); 4.56 (dd, 1H, J 10.5 Hz, 11.8 Hz, H2), 4.75 (dd, 1H, J 1.3 Hz, 3.2 Hz, H4), 6.66 (dd, 1H, J 2.3 Hz, 8.7 Hz, H6), 6.69 (d, 1H, J 2.3 Hz, H8), 7.08 (d, 2H, J 8.6 Hz, ArH), 7.26 (d, 1H, 8.4 Hz, H5), 7.29 (d, 2H, J 8.6 Hz ArH). ^{13}C NMR ($CDCl_3$); δ 20.98 ($OCOCH_3$), 43.52 (C3), 64.10 (C2), 66.46 (C4), 110.08 (C6), 114.09 (C8), 121.82, 129.40 (ArCH), 131.10 (C5).

EXAMPLE 4

Synthesis of 7-Acetoxy-4'-methoxyisoflavan-4-ol

Palladium-on-charcoal (5%, 0.08g) was added to a suspension of 7-acetoxy-4'-methoxyisoflavanone (0.5g, 1.6 mmol) in absolute ethanol (400 ml) and the mixture was stirred at room temperature under a hydrogen atmosphere for 55 hours. The catalyst was removed by filtration through Celite and the filtrate was evaporated *in vacuo* to yield 7-acetoxy-4'-methoxyisoflavan-4-ol (0.51g, 100%) in quantitative yield. A nuclear magnetic resonance spectrum revealed the product to be a clean 1:1 mixture of *cis* and *trans* 7-acetoxy-4'-methoxyisoflavan-4-ol.

trans-7-Acetoxy-4'-methoxyisoflavan-4-ol; 1H NMR ($CDC1_3$): d 2.31 (s, 3H, $OCOCH_3$), 3.14 (dt, 1H, J 3.8 Hz, 8.6 Hz, H3), 2.29 (s, 3H, $OCOCH_3$), 3.82 (s, 3H, OCH_3), 4.25 (dd, 1H, J 9.4 Hz, 11.3 Hz, H2); 4.37 (dd, 1H, J 4.1 Hz, 11.3 Hz, H2), 4.93 (d, 1H, J 7.8 Hz, H4), 6.63 (d, 1H, J 2.3 Hz, H8), 6.73 (dd, 1H, J 2.3 Hz, 8.3 Hz, H6), 6.93 (d, 2H, J 8.7 Hz, ArH), 7.19 (d, 2H, J 8.7 Hz, ArH), 7.51 (d, 1H, J 7.9 Hz, H5).

cis-7-Acetoxy-4'-methoxyisoflavan-4-ol; 1H NMR ($CDC1_3$): d 2.30 (s, 3H, $OCOCH_3$), 3.28 (dt, 1H, J 3.4 Hz, J 12.1 Hz, H3), 3.84 (s, 3H, OCH3), 4.36 (ddd, 1H, J 1.4 Hz, 3.8 Hz, 10.1 Hz, H2); 4.57 (dd, 1H, J 10.1 Hz, 11.3 Hz, H2), 4.75 (bs, 1H, H4), 6.58 (d, 1H, J 2.3 Hz, H8), 6.75 (dd, 1H, J 2.3 Hz, 8.3 Hz, H6), 6.96 (d, 2H, J 8.6 Hz, ArH), 7.25 (d, 2H, 8.6 Hz, ArH), 7.34 (d, 1H, J 8.3 Hz, H5).

EXAMPLE 5

Separation of *cis*- and *trans*-4',7-diacetoxytetrahydrodaidzein

A 1 : 1 mixture of cis and trans 4',7-diacetoxytetrahydrodaidzein (0.17 g), prepared as above, was dissolved in excess absolute ethanol and concentrated on a rotary evaporator. At the first sign of crystallisation, further concentration of ethanol was stopped and the flask was cooled in an ice-bath. The resulting crystals were filtered and washed with a small amount of cold absolute ethanol. A nuclear magnetic resonance spectrum of the product (0.08 g) revealed it to be a mixture trans-4',7-diacetoxytetrahydrodaidzein (73%) and cis-4',7-diacetoxytetrahydrodaidzein (27%). Further recrystallisations of the mixture from ethanol yielded the pure trans-4',7-diacetoxytetrahydrodaidzein (0.04 g, 24%).

The filtrate yielded predominantly cis isomer. Nuclear magnetic resonance spectroscopic analysis revealed the substance to be a mixture of cis-4',7-diacetoxytetrahydrodaidzein (73%) and trans-4',7-diacetoxytetrahydrodaidzein (27%).

EXAMPLE 6

Separation of *cis*- and *trans*-7-acetoxy-4'-methoxyisoflavan-4-ol

A 1:1 mixture of *cis* and *trans* 4',7-diacetoxytetrahydrodaidzein prepared as above was recrystallised three times from ethanol to yield pure *trans*-7-acetoxy-4'-methoxyisoflavan-4-ol. The filtrate yielded predominantly *cis* isomer.

EXAMPLE 7

***cis*- and *trans*-Tetrahydrodaidzein**

Imidazole (0.2 g) was added to a suspension of 4',7-diacetoxydihyrodaidzein (0.10 g, 0.3 mmol) in absolute ethanol (4.0 ml) and the mixture refluxed for 45 min under argon. The solution was concentrated under reduced pressure and distilled water (10 ml) was added. The mixture was left overnight in the fridge and the crystalline product was filtered to yield cis- and trans-tetrahydrodaidzein (0.06 g, 80%).

EXAMPLE 8

***cis*- and *trans*-7-Hydroxy-4'-methoxyisoflavan-4-ol**

Imidazole (0.4 g) was added to a suspension of 7-acetoxy-4'-methoxyisoflavan-4-ol (0.20 g, 0.6 mmol) in absolute ethanol (8.0 ml) and the mixture refluxed for 45 minutes under argon. The solution was concentrated under reduced pressure and distilled water (10 ml) was added. The mixture was left overnight in the fridge and the crystalline product was filtered to yield *cis*- and *trans*-7-hydroxy-4'-methoxyisoflavan-4-ol (0.16g, 79%).

EXAMPLE 9

4',7-Diacetoxydehydroequol (4',7-Diacetoxyisoflav-3-ene)

Method A

Distilled trifluoroacetic acid (0.1 ml) was added to a solution of *cis*- and *trans*-4',7-diacetoxytetrahydrodaidzein (0.1g) in dry distilled dichloromethane (15 ml) and the mixture was refluxed under argon. Progress of the reaction was monitored by thin layer chromatography and further 0.1 ml portions of trifluoroacetic acid were added. After refluxing for 4 hours, the reaction mixture was cooled and washed successively with saturated sodium bicarbonate solution, water and brine. The resulting organic phase was dried, concentrated, chromatographed and crystallised to yield 4',7-diacetoxydehydroequol as colourless prisms (0.034g, 35%). ^{1}H NMR ($CDCl_3$ + d_6-DMSO): δ 2.29 (s, 3H, $OCOCH_3$), 2.31 (s, 3H, $OCOCH_3$), 5.15 (s, 2H, H2), 6.62 (bs, 1H, H4), 6.65 (dd, 1H, J 2.1 Hz 8.2 Hz, H6), 6.75 (bs, 1H, H8), 7.06 (d, 1H, J 8.2 Hz H5), 7.12 (d, 2H, J 8.2 Hz, ArH), 7.43 (d, 2H, J 8.2 Hz, ArH).

Method B

p-Toluenesulfonic acid (0.02 g) was added to a solution of cis- and trans-4'7-diacetoxytetrahydrodaidzein (0.1 g) in dry distilled dichloromethane (15 ml) and the mixture was refluxed under argon. Progress of the reaction was monitored by thin layer chromatography and after 4h at reflux, the reaction mixture was passed through a short column of silica gel and the eluant recrystallised from ethanol to yield 4',7-diacetoxydehydroequol as colourless prisms (0.025 g, 26%).

EXAMPLE 10

7-Acetoxy-4'-methoxyisoflav-3-ene

Phosphorus pentoxide (1.0g) was added with stirring to a solution of *cis*- and *trans*-7-acetoxy-4'-methoxyisoflavan-4-ol (0.1g, 0.3 mmol) in dry toluene (20 ml). The mixture was heated under reflux for 2 hours. After cooling to room temperature, the mixture was treated carefully with water (40 ml). The organic phase was washed with water (50 ml) followed by brine (50 ml). The resulting solution was dried over sodium sulfate, concentrated and chromatographed on silica gel to yield 7-acetoxy-4'-methoxyisoflav-3-ene (0.04g, 42%). 1H NMR ($CDC1_3$); d 2.28 (s, 3H, OCOCH3), 3.83 (s, 3H, OCH3), 5.14 (s, 2H, H2), 6.61 (dd, 1H, J 2.3 Hz 6.4 Hz, H6), 6.65 (d, 1H, J 2.3 Hz, H8), 6.69 (bs, 1H, H4), 6.92 (d, 2H, J 9.0 Hz ArH), 7.04 (d, 1H, J 7.9 Hz, H5), 7.37 (d, 2H, J 9.0 Hz, ArH).

EXAMPLE 11

Dehydroequol (Isoflav-3-ene-4',7-diol)

Imidazole (0.09 g) was added to a suspension of 4',7-diacetoxydehydroequol (0.03 g, 0.09 mmol) in absolute ethanol (2.0 ml) and the mixture was refluxed for 45 min under argon. The solution was concentrated under reduced pressure and the product was precipitated by addition of distilled water (10 ml). The mixture was left overnight in the fridge and filtered to yield dehydroequol. The crude product was reprecipitated from methanol by addition of benzene to yield dehydroequol as fluffy white solid (0.012 g, 55%). ^{1}H NMR ($CDCl_3$ + d_6-DMSO): δ 4.93 (s, 2H, H2), 6.26 (bs, 1H, H4), 6.29 (dd, 1H, J 2.0 Hz, 8.2 Hz, H6), 6.50 (bs, 1H, H8), 6.73 (d, 2H, J 8.2 Hz, ArH), 6.76 (d, 2H, J 8.2 Hz, H5), 7.13 (d, 2H, J 8.2 Hz, ArH).

EXAMPLE 12

7-Hydroxy-4'-methoxyisoflav-3-ene

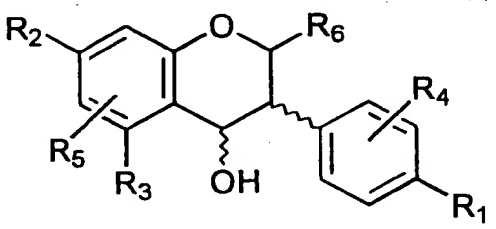
Imidazole (0.18g) was added to a suspension of 7-acetoxy-4'-methoxyisoflav-3-ene (0.06g, 0.02 mmol) in absolute ethanol (5.0 ml) and the mixture was refluxed for 45 minutes under argon. The solution was concentrated under reduced pressure and the product was precipitated by addition of distilled water (10 ml). The mixture was left

overnight in the fridge and filtered to yield isoflav-3-ene. The crude product was recrystallised from methanol/benzene to yield 7-hydroxy-4'-methoxyisoflav-3-ene (0.034g, 66%). 1H NMR ($CDCl_3$ + d_6-DMSO): d 3.74 (s, 3H, OCH_3), 4.99 (s, 2H, H2), 6.21 (d, 1H, J 2.3 Hz, H8), 6.29 (dd, 1H, J 2.3 Hz, 8.3 Hz, H6), 6.67 (bs, 1H, H4), 6.85 (d, 1H, J 8.3 Hz, H5), 6.86 (d, 2H, J 8.7 Hz, ArH), 7.33 (d, 2H, J 8.7 Hz, ArH).

Those skilled in the art will appreciate that the invention described herein is susceptible to variations and modifications other than those specifically described. It is to be understood that the invention includes all such variations and modifications. The inventions also includes all of the steps, features, compositions and compounds referred to or indicated in the specification, individually or collectively, and any and all combinations of any two or more of said steps or features.

The claims defining the invention are as follows:

1. A method for the preparation of a compound of formula II



(II)

wherein

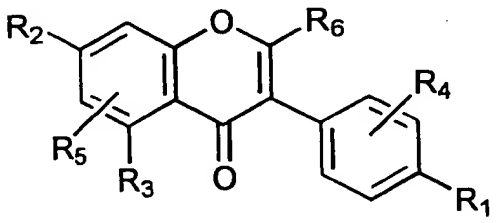
R_1 and R_2 are independently OR_7, $OC(O)R_7$, $OS(O)R_7$, alkyl, aryl, arylalkyl, thio, alkylthio, amino, alkylamino, dialkylamino, nitro, or halo,

R_3, R_4, R_5 and R_6 are independently hydrogen, hydroxy or R_1, and

R_7 is alkyl, fluoroalkyl, aryl or arylalkyl,

to prepare a compound of formula II

comprising the step of hydrogenating a compound of formula I



(I)

wherein

R_1, R_2, R_3, R_4, R_5, R_6 and R_7 are as defined above.

2. A method of claim 1, wherein the hydrogenation step is performed with hydrogen in the presence of a reduction catalyst and a solvent.

3. A method of claim 2, wherein the reduction catalyst is palladium, palladium hydroxide, platinum or platinum oxide.

4. A method of claim 3, wherein the reduction catalyst is palladium on activated carbon or palladium on barium sulfate.

5. A method of claim 4, wherein the reduction catalyst is palladium on activated carbon (1% Pd to 10% Pd).

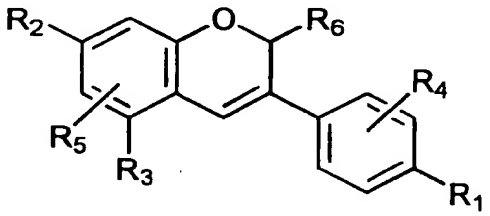
6. A method of claim 5, wherein the reduction catalyst is about 5% palladium on activated carbon.

7. A method of claim 2, wherein the solvent is a C_1-C_8 alcohol or polyol, an alkyl acetate, tetrahydrofuran, an ether, dioxane or a C_1-C_3 carboxylic acid.

8. A method of claim 7, wherein the solvent is a C_1-C_6 alcohol or C_1-C_6 alkyl acetate.

9. A method of claim 8, wherein the solvent is absolute ethanol.

10. A method of claim 1 which further comprises the step of dehydrating and optionally deprotecting or transforming a compound of formula II to prepare a compound of formula III



(III)

wherein

R_1 and R_2 are independently hydroxy, OR_7, $OC(O)R_7$, $OS(O)R_7$, alkyl, aryl, arylalkyl, thio, alkylthio, amino, alkylamino, dialkylamino, nitro, or halo,

R_3, R_4, R_5 and R_6 are independently hydrogen, hydroxy or R_1, and

R_7 is alkyl, fluoroalkyl, aryl or arylalkyl.

11. A method of any one of claims 1 to 10, wherein the compounds of formula I, II or III have the following substituents
R_1 and R_2 are independently OR_7, $OC(O)R_7$, $OS(O)R_7$, thio, alkylthio, bromo, chloro or fluoro,
R_3, R_4, R_5 and R_6 are independently hydrogen or R_1, and
R_7 is alkyl or fluoroalkyl.

12. A method of claim 11, wherein the compounds of formula I, II or III have the following substituents
R_1 and R_2 are independently OR_7 or $OC(O)R_7$,
R_3, R_4, R_5 and R_6 are hydrogen, and
R_7 is methyl, ethyl, propyl, isopropyl or trifluoromethyl.

13. A method of claim 12, wherein the compounds of formula I, II or III have the following substituents
R_1 and R_2 are independently OR_7 or $OC(O)R_7$,
R_3, R_4, R_5 and R_6 are hydrogen, and
R_7 is methyl or trifluoromethyl.

14. A method of any one of claims 1 to 10, wherein the compound of formula I is 4',7-diacetoxyisoflavanone (daidzein diacetate) or 7-acetoxy-4'-methoxyisoflavone, the compound of formula II is 4',7-diacetoxyisoflavan-4-ol (tetrahydrodaidzein diacetate) or 7-acetoxy-4'-methoxyisoflavan-4-ol and the compound of formula III is 4',7-diacetoxyisoflav-3-ene (dehydroequol diacetate), 4',7-dihydroxyisoflav-3-ene (dehydroequol), 7-acetoxy-4'-methoxyisoflav-3-ene or 7-hydroxy-4'-methoxyisoflav-3-ene.

15. Methods substantially as hereinbefore described with reference to the Examples.

16. Compounds of formula II or formula III when prepared by a method of any preceding claim.

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NOVOGEN RESEARCH PTY LTD

By its Patent Attorneys
DAVIES COLLISON CAVE